



First record of *Chlorophyllum molybdites* (G. Mey.) Massee (Basidiomycota, Agaricales, Agaricaceae) from Bucaramanga, Department of Santander, Colombia

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Abstract

Colombia is a megadiverse country with many macrofungi species. Despite their ecological, anthropological, and economic importance, the distribution of many macrofungal species is still scarcely known in this country. *Chlorophyllum molybdites* (G. Mey.) Massee is a poisonous macrofungus common in urban settings of tropical and subtropical regions. In Colombia it has only been reported in three of 32 departments. To fill this gap in knowledge we morphologically describe for the first time *C. molybdites* from the city of Bucaramanga, Department of Santander, extending the known distribution of this macrofungus in Colombia.

Keywords

Biodiversity, geographical distribution, gilled mushroom, macrofungi, South America, Tropics, urban species

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Introduction

Macrofungi include ascomycetes and basidiomycetes with large, easily observed fruiting bodies. They are an ecologically important group of organisms due to their essential roles as decomposers of organic material, in mutualistic associations with plants, and as parasites or pathogens of other fungi, plants, and animals (Pérez-Moreno et al 2020). In a developing country such as Colombia, macrofungi not only fulfill ecologically important roles but are also crucial for certain anthropological and economical activities. For example, the cultivation of edible macrofungi is an important economic

activity that generates income for marginalized groups (Vargas et al. 2020) and a biotechnological alternative to obtain compounds of medical use (Suárez-Arango and Nieto 2013). Therefore, surveying and monitoring macrofungi distribution and biodiversity in this developing country is essential to support ecological, biotechnological, and ethnobiological research.

Colombia is a megadiverse country thanks to its position in the Equatorial region (Fig. 1A) and its complex geography. Although the number of studies on Colombia's biodiversity has been increasing, studies on fungal

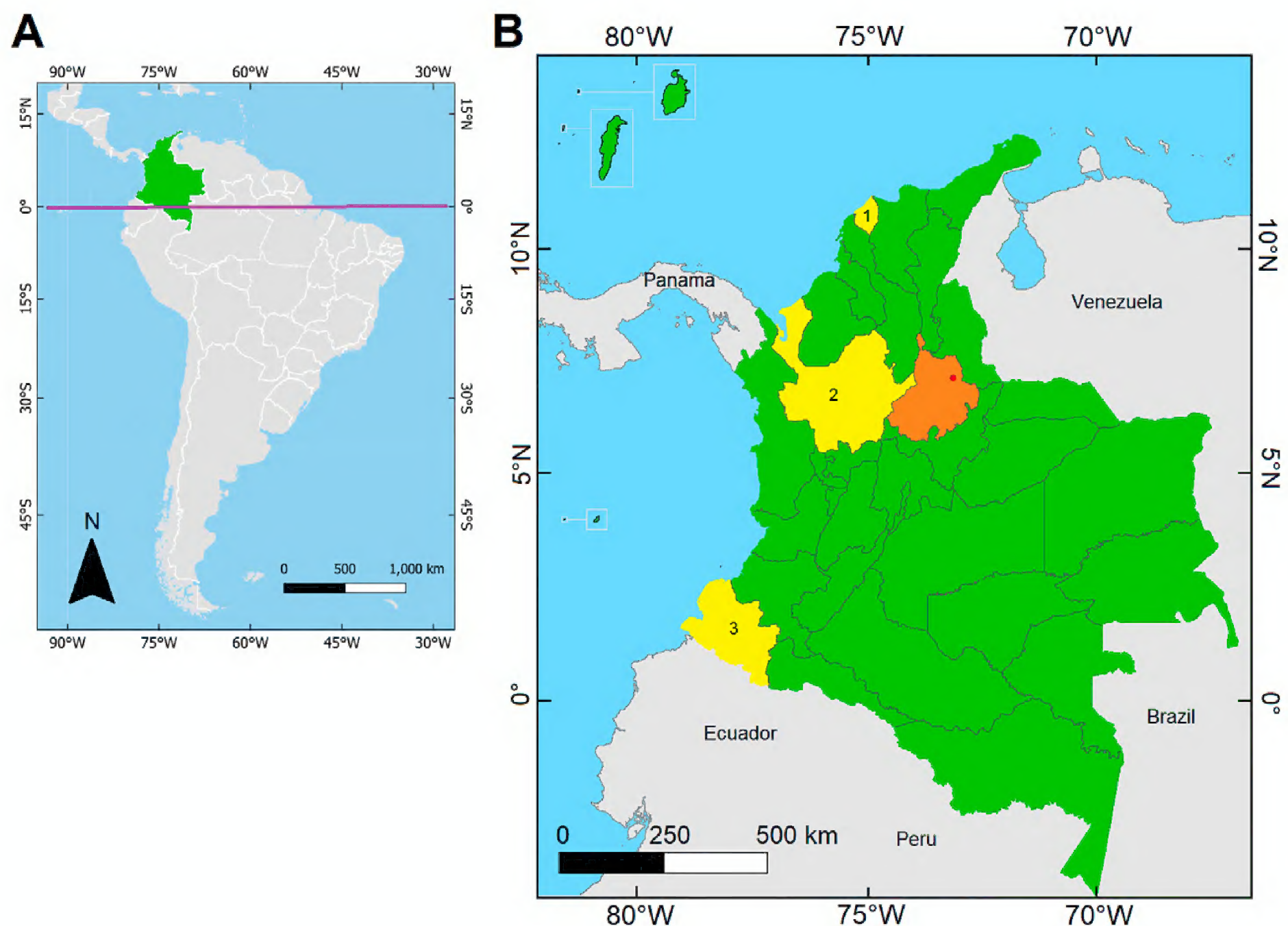


Figure 1. Maps of South America and Colombia. **A.** Map of South America in which Colombia is shown in green and the Equator line is in violet. **B.** Map of Colombia. The 32 departments of Colombia are shown. The three departments where *Chlorophyllum molybdites* has been previously reported are shown in yellow: 1 = Magdalena, 2 = Antioquia, 3 = Nariño. This paper reports *C. molybdites* for the first time in the Department of Santander (in orange) and the collection site, the city of Bucaramanga, is represented as a red dot.

biodiversity are scarce when compared to other taxa (Arbeláez-Cortés 2013). However, a systematic study of Colombian macrofungi reported a staggering 1239 species, with more than 85% of those reported belonging to Basidiomycota (Vasco-Palacios and Franco-Molano 2013). Despite this incredible diversity, surveys on the distribution of macrofungi in Colombia are geographically biased (Franco-Molano et al. 2010). For example, Colombia is politically divided into 32 departments, and only in 24 of these have macrofungal collections been reported (Vasco-Palacios and Franco-Molano 2013). Therefore, descriptive studies that document the presence of macrofungi in neglected regions of Colombia are important to explore macrofungi distribution at the local scale. One of these neglected regions is the Department of Santander in which only 23 species of macrofungi have been reported, a small number compared to more than 300 reported in other well-studied departments (Vasco-Palacios and Franco-Molano 2013).

Chlorophyllum molybdites (G. Mey.) Masee is a poisonous macrofungus belonging to the order Agaricales, family Agaricaceae (Vellinga et al. 2003). In South America, it has been reported in Argentina (Romano et al. 2013), Brazil (Guzmán 1986; Sobestiansky 2005; Remi de Meijer et al. 2007; Drechsler-Santos 2008;

Somenzi Rother and Borges da Silveira 2008; Alves et al. 2016; Alves et al. 2019), Colombia (Franco-Molano et al. 2000; Franco-Molano et al. 2010; Vasco-Palacios and Franco-Molano 2013), French Guiana (Pegler 1983; Guzmán 1986), Guyana (Pegler 1983; Guzmán 1986), Trinidad (Dennis 1952; Pegler 1983), and Venezuela (Dennis 1970). It has been reported as the most common cause of mushroom poisoning in Argentina (Romano et al. 2013), and in Brazil severe gastrointestinal symptoms have been reported after its consumption; these symptoms include vomiting, abdominal pain, overall weakness, and diarrhea (Remi de Meijer et al. 2007). As in North America (Blayney et al. 1980), poisoning in South America seems to be due to the misidentification of this mushroom for edible species which in South America include *Macrolepiota bonaerensis* (Speg.) Singer and *Chlorophyllum hortense* (Murrill) Vellinga (Remi de Meijer et al. 2007). *Chlorophyllum molybdites* has only been reported from three Colombian departments (Fig. 1B): Antioquia, Magdalena, and Nariño (Franco-Molano et al. 2000, 2010; Vasco-Palacios and Franco-Molano 2013). As *C. hortense* has also been documented in Colombia (Luna-Fontalvo et al. 2021) and *C. molybdites* is common in lawns and other environments manipulated or transformed by humans in

tropical and subtropical areas (Vellinga 2003), its correct identification is important for preventing possible poisoning in humans. In this study we morphologically describe the first specimens of *C. molybdites* from the urban setting of the city of Bucaramanga, the most populated city of the Department of Santander (Fig. 1B), and we extend the known distribution of this macrofungus in Colombia.

Methods

At the beginning of April 2019 several basidiocarps with typical morphological features of agaricoid mushrooms of large size were observed in the District of Mejoras Públicas, Commune 13, in the city of Bucaramanga, Department of Santander, Colombia (Fig. 1B). The basidiocarps were found in a small grassy area that separates a cycling path from a busy street. Planted Italian cypress trees (*Cupressus sempervirens* L.) were also found in the grassy area. Photographs of the individuals were taken on site with a digital camera, and five basidiocarps were collected on 5 April 2019, using wax paper to wrap each individual. The colors of each macroscopic feature were recorded using the CMYK color model, following the Pantone Plus Series CMYK Guide set GP5101. This color guide set is based on the concept that all colors are a combination of four basic colors: Cyan (C), Magenta (M), Yellow (Y) and Black (K). Therefore, each color is represented by four numbers separated by commas which indicate the color values of C, M, Y and K, respectively.

Macroscopic features were recorded immediately after collection following Largent (1986). Afterwards, spore prints were obtained of each individual on white paper. All individuals were dried overnight in an incubator at 65 °C. For the documentation of microscopic features, dried tissues were rehydrated using 3% KOH. Tissues and spores were stained with a 1% Congo red aqueous solution and Melzer's reagent and observed using an Axio Observed 5 inverted-phase contrast microscope (Carl Zeiss). Photographs were taken using an AxioCam 503 color (Carl Zeiss) attached to the microscope. For each of the five specimens collected, 20 spores, 10 basidia, and 10 cheilocystidia were measured using the Zen 2.5 Pro (blue edition) software (Carl Zeiss). We followed the key to the species of *Chlorophyllum* by Ge et al (2018) in which the macroscopic and microscopic features of the collected specimens are used to identify the species to which the specimens belong.

Results

Chlorophyllum molybdites (G. Mey.) Massee

New record. COLOMBIA – Department of Santander • Bucaramanga, Commune 13, District of Mejoras Públicas; 07°07'34"N, 073°06'54"W; 1013 m above sea level; 5.IV.2019; X.A. Olarte-Castillo leg.; XOC1-5.

Identification. Macroscopic description. For each macroscopic feature the smallest and largest values measured are reported. Pileus (Fig. 2A–C) 4.5–22 cm in diameter, ovoid when young, plane to convex when old, some old specimens may be umbonated, decurved margin, squamose surface, surface dull and cream colored (0,2,10,0), turns dark cream (2,5,10,0) to light brown (20,30,30,0) with age, in some specimens the surface is light brown (20,30,30,0) in the center of the cap, a large scale remains in the center of the pileus that may or may not break, scales are raised, cream-colored (0,5,10,0) to light brown (15,20,30,0) when young then turn dark brown (35,50,70,0) with age, towards the margin the surface may be smooth or scaly and scales remain cream-colored (2,5,10,0), striate margin. Context dry, light cream (2,5,10,0) to dark cream (5,5,10,0) that may be darker than the pileus color, 0.2–1.2 cm towards the disc, 0.1–0.2 cm towards the margin, spongy texture. Lamellae (Fig. 2D) free and remote, narrow to broad, closely spaced, of average thickness, margin even to eroded, unequal due to the presence of lamellulae, 0.2–1.2 cm, cream-colored (2,5,10,0) when young, greenish (35,25,50,0) with age due to the spores. Stem (Fig. 2D, E) central, cream-colored (2,5,10,0) when young, light brown (15,20,30,0) when old, 5–21 cm in height, 0.5–1.3 cm in diameter, flexous, longitudinally striate, flexible, interior hollow, interior cream-colored (5,5,10,0), lighter than the stem. Partial veil white (0,0,10,0), cortinate, sticky, disappearing leaving white (0,0,10,0) threads on the grass and a ring. Annulus (Fig. 2C, D) single, thick on edge, superior, cream-colored (2,5,10,0) when young but darker 5,5,10,0) with age. Spore print (Fig. 2F) grayish green 35,25,50,0) to olive-green (20,15,30,0). Non distinctive taste, mushroom-like odor when dried. In total 15 basidiocarps with similar macroscopic features were observed in the same grassy area from 3 April to 12 May 2019. These were scattered to gregarious with mostly two individuals growing very close to each other (Fig. 2B). Many grew close to the planted Italian cypress trees.

Microscopic description. The size of each microscopic structure is reported as mean and standard deviation. Basidia (Fig. 3A, B) 25.8 (± 3.7) µm, 12.1 (± 2.0) µm (length, width), tetraspored, clavate to broadly clavate, hyaline. Basidiospores (Fig. 3C) 8.2 (± 0.5) µm, 6.8 (± 1.2) µm (length, width), ellipsoid to amygdaliform with germ pore, smooth, with a thick wall, dextrinoid in Melzer's reagent. Pleurocystidia absent. Cheilocystidia (Fig. 3D) 28.6 (± 2.3) µm, 12.3 (± 1.4) µm (length, width), hyaline, broadly clavate to sphaero-pedunculate, wall smooth and thin.

Discussion

In this study we observed several basidiocarps, recorded their macroscopic (Fig. 2) and microscopic (Fig. 3) morphological features, and followed the key by Ge et al. (2018). We concluded that this macrofungus belongs to the species *Chlorophyllum molybdites*. Although

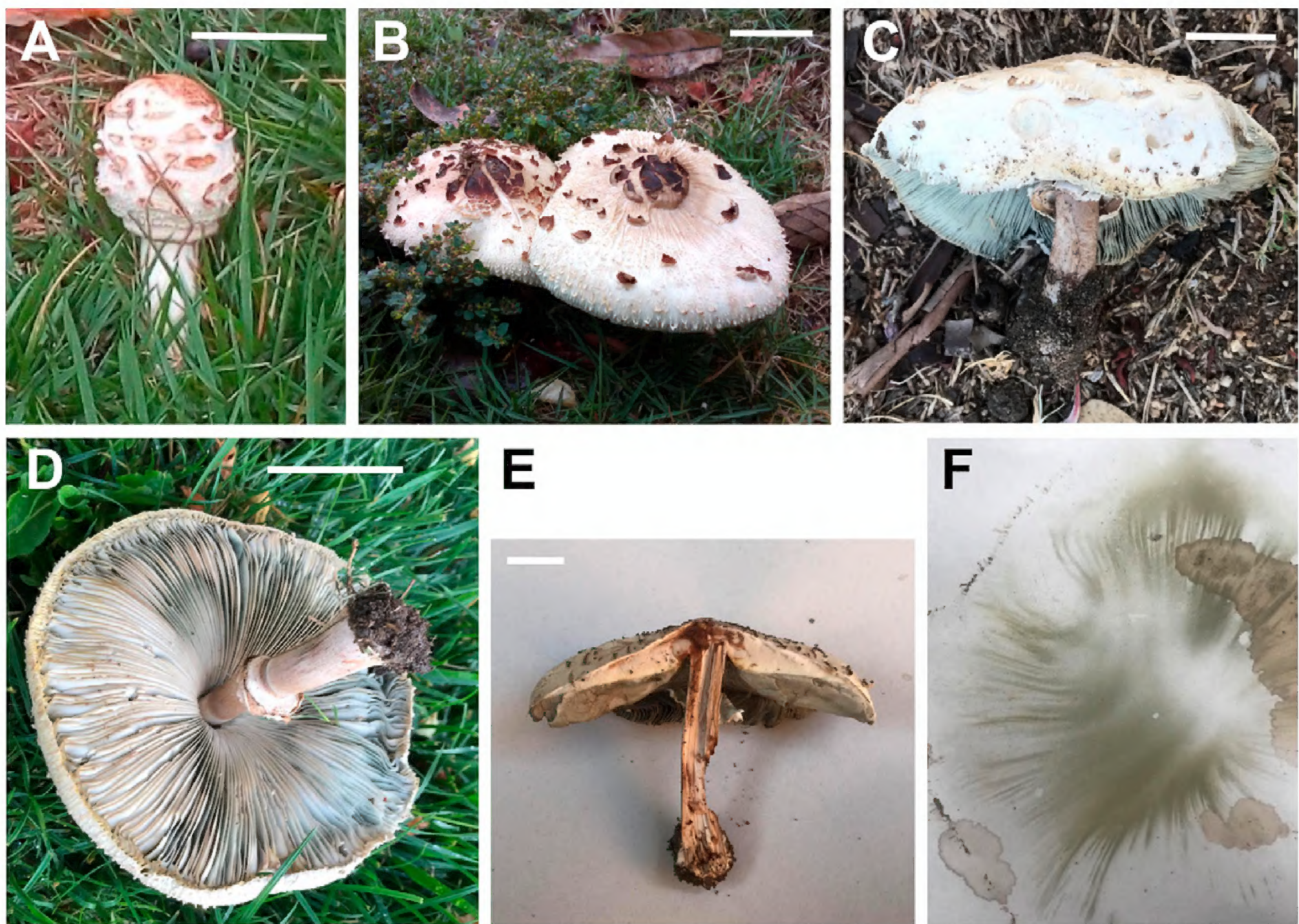


Figure 2. Macroscopic features of *Chlorophyllum molybdites*. **A.** Side view of the basidiocarp of a young individual. **B.** Upper view of the basidiocarp in which the cream-colored cap with brown and cream-colored scales are shown. **C.** Side view of the basidiocarp in which the cap, stipe, gills, and ring are shown. **D.** Lower view of the basidiocarp in which green and cream-colored gills, stipe and ring are shown. **E.** Cross section of an individual showing the context, free and remote lamellae, and hollow stipe. **F.** Green spore print. Scale bars: A–E = 5 cm.

molecular methods can be used in the identification of this species (Wang et al. 2021), *C. molybdites* has several key morphological attributes that make it distinct from other mushroom species. The observed specimens had all these characteristics including cream-colored agaricoid basidiocarps growing in grass (Fig. 2B), brown scales on the pileus (Fig. 2A), basidiospores with germ pore (Fig. 3C), a representative green spore print (Fig. 2F), and lamellae that turn completely greenish with age (Fig. 2D). Therefore, in this study we describe for the first time *C. molybdites* from the city of Bucaramanga, in the Department of Santander (Fig. 1B) and extend the known distribution of this species to a region of Colombia where macrofungal biodiversity is poorly explored (Vasco-Palacios and Franco-Molano 2013).

Chlorophyllum molybdites is commonly reported from urban settings, where it can be one of the most commonly encountered macrofungus species (Esqueda-Valle et al. 1995). We report this species in the city of Bucaramanga, the most populated municipality in the Department of Santander (DANE 2020). We found the specimens in a small patch of grass next to a busy street showing that this species can thrive in a highly disturbed area. In Medellin, Department of Antioquia (Fig. 1B), another highly populated city of Colombia

and in southern Brazil, it has also been reported in similar urban areas (Somenzi Rother and Borges da Silveira 2008; Lopez-Quintero et al. 2011). As this species is poisonous and readily reported from urban settings, its rapid morphological identification is important to avoid confusing it with other edible species (Romano et al. 2013).

We observed basidiocarps between 3 April and 12 May 2019, which coincides with the first rainy season of the year in this area (IDEAM 2010). In other South American cities, this species has also been reported during the rainy season (Lopez-Quintero et al. 2011; Alves et al. 2019). We report this species from a lower elevation (1013 m above sea level) than previously known in Colombia (1450–1550 m above sea level: Vasco-Palacios and Franco-Molano 2013) and in a city with a warmer average temperature (22.6 °C: IDEAM 2010) than another city where this species has been previously reported in this country (20 °C: Lopez-Quintero et al. 2011).

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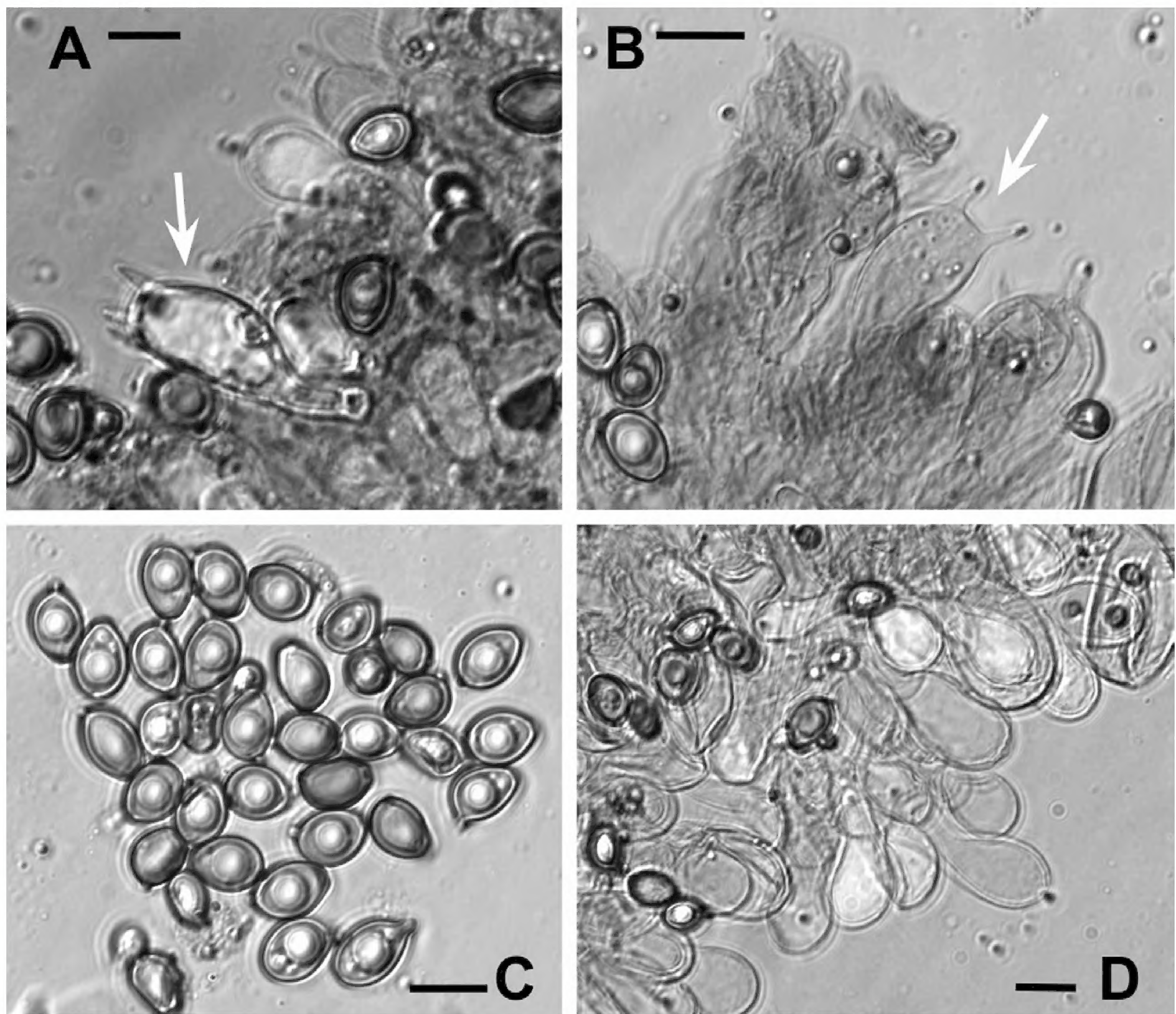


Figure 3. Microscopic features of *Chlorophyllum molybdites*. **A, B.** Basidia shown with a white arrow. **C.** Basidiospores. **D.** Cheilocystidia. Scale bars = 10µm.

Authors' Contributions

Conceptualization: XAOC. Data curation: XAOC, CAA. Formal analysis: CAA. Funding acquisition: CAA. Investigation: XAOC. Methodology: CAA. Resources: XAOC, CAA. Visualization: XAOC. Writing – original draft: XAOC. Writing – review and editing: XAOC.

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